



PRESIDENT'S MALARIA INITIATIVE



Advanced Entomology Technicians Training Report—Liberia

Integrated Vector Management (IVM) Task Order 2

Contract GHA-I-02-04-00007-00

Prepared for:

United States Agency for International Development

Prepared by:

RTI International
3040 Cornwallis Road

Post Office Box 12194
Research Triangle Park, NC 27709-2194

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November 2010

This report was produced for review by the United States Agency for International Development. It was prepared by RTI International. The author's views expressed in this report do not necessarily reflect the views of the United States Agency for International Development or the United States Government.

Consultants Report on Intermediate Entomology Technician's Training Course

23rd August – 22nd October 2010

Noguchi Memorial Institute for Medical Research

Accra, Ghana

Consultants:

Dr Maxwell Appawu (Medical Entomologist)
Noguchi Memorial Institute for Medical Research,
P. O. Box LG581, Legon, Ghana
Email: mappawu@noguchi.mimcom.org

Prof. Daniel Boakye (Medical Entomologist)
Noguchi Memorial Institute for Medical Research,
P. O. Box LG581, Legon, Ghana
Email: dboakye@noguchi.mimcom.org

Dr Samuel Dadzie (Medical Entomologist)
Noguchi Memorial Institute for Medical Research,
P. O. Box LG581, Legon, Ghana
Email: sdadzie@noguchi.mimcom.org

1.0 Introduction

The workshop on advanced intermediate entomology training was organized at Noguchi Memorial Institute for Medical Research (NMIMR) in Ghana. The preparation of course materials began on the 19th of August 2010. The course commenced on the 23rd of August and ended on 22nd of October 2010. The course was funded by Research Triangle International (RTI) in Washington, USA.

Four (4) participants from Liberia took part in the course. They were Chrispin Williams and George K. Gweh from Liberia National Malaria Control Programme; Lawrence S. Fakoli III and William T. Gayflor I from the Liberia Institute for Biomedical Research).

The facilitators for the course were Dr. Maxwell Appawu, Prof. Daniel Boakye and Dr. Samuel Dadzie (Noguchi Memorial Institute for Medical Research, Ghana), with support from Kathryn Welter (RTI International, Washington, USA).

The overall aim of the course was to build capacity in entomological monitoring of vector control programmes in Liberia and help strengthen the national vector control in the country.

Broadly, the course was aimed at equipping participants with techniques such as:

- Advanced mosquito sample preparations: preparation/preservation for Enzyme Linked Immunosorbent Assay (ELISA)/Polymerase Chain Reaction (PCR), dissection (ovaries and salivary gland), parity evaluation, etc.
- ELISA-based entomology techniques to support monitoring: sporozoite detection, blood meal analysis, etc.
- In-depth review of mosquito ecology and their control, including advanced sampling techniques, mosquito rearing techniques (in-depth), and considerations on sentinel monitoring and surveillance (in-depth).

At the end of the course each participant was to be able to:

- understand the principles and carry out advanced vector sampling techniques
- understand mosquito ecology and how it impacts on the formulation of vector control.

- To do mosquito sample preparation and preservation for ELISA or PCR.
- dissect ovaries and salivary glands of mosquitoes for *Plasmodium* infectivity and parity evaluation.
- carry out ELISA techniques for sporozoite and blood meal analysis.
- undertake mosquito rearing and maintenance in the insectary.
- analyse and interpret entomological data.

The course was participatory and interactive and to ensure relevance of course, each participants took a pre- and post course examination. There were also extensive field and laboratory work as well as practical demonstrations.

2.0 Lectures

To enable the participants understand the principles and theory behind the entomological techniques, the participants were taken through intensive lectures with topics on mosquito ecology, transmission dynamics and control; principles of Enzyme Linked Immunorsorbent Assay, microscopy, adult mosquito and larval sampling, mosquito rearing techniques (Annex 1). Lectures were given on the following topics :

- 1) The objectives of the course
 - *The objectives and course structure*
- 2) The Biology of Malaria Vectors
 - *Mosquito life cycle and the significance of each stage of life cycle in malaria control, vector behaviour and its significance in control*
- 3) Mosquito Anatomy and Identification
 - *External morphological diagnostic features of different species of mosquitoes and how to use the features for identification*
- 4) Mosquito survey - Methods for sampling larvae
 - *The participants were taught the different sampling techniques for collecting larvae and the advantages and disadvantages of each method*

- 5) Vector incrimination
 - *Factors such as mosquito abundance, longevity, susceptibility to infection and anthropophily that determines a mosquito as vector*
- 6) Adult Mosquito Survey
 - *Adult mosquito sampling techniques such as landing catches, pyrethrum spray collection, outdoor collections, exit trap collection and hand aspirator collection*
- 7) Determination of transmission parameters
 - *Estimation of malaria transmission parameters such as man biting rates, sporozoite rates, parity rates and Entomological inoculation rates*
- 8) Integrated Vector Management (IVM)
 - *The principle and concept of IVM*
- 9) Susceptibility/Resistance of mosquitoes to insecticides
 - *Resistance mechanisms and methods to detect resistance in mosquitoes, implications of the development of resistance in mosquitoes*
- 10) Insectary Management
 - *Breeding and maintenance of mosquito colonies using Standard Operating Procedures for insectary)*
- 11) Enzyme Linked Immunosorbent assay (ELISA)
 - *Principles of ELISA including antibody and antigen reactions, enzyme and substrate reactions*
- 12) Malaria Vector Control
 - *Principles and concepts behind various malaria control methods and the advantages and disadvantages of each method*

Two videos on Field Entomological Techniques and insecticide bioassays were shown to the participants several times during the duration of the training.

3.0 Laboratory work

The participants were initially taken through basic laboratory techniques such as microscopy, pipetting and standard operating procedures in the laboratory. Each participant was taken through mosquito identification, sample preservation and preparation for ELISA and PCR, dissections for parity evaluation, ELISA technique for detection of *Plasmodium* species and source of blood meal in mosquitoes as well as WHO methods for detecting insecticide resistance. The participants were taken through the use of microplate ELISA reader and how to analyse data after running samples in the reader (Annex 1). Initially, the participants worked in groups of two to emphasize the importance of team work but later each participant was allowed to work independently under supervision from experienced research assistants and facilitators.

3.1 Mosquito identification

Participants were taught how to use some morphological features of mosquitoes to sort out mosquitoes into different species (Picture 1). Exercises on how to calculate man-biting rates, sporozoite rates and entomological inoculation rates were carried out.



Picture 1: Participants sorting and identifying mosquitoes from the field

3.2 Participants doing ELISA technique

The participants were taken through ELISA techniques for detecting Plasmodium parasite and bloodmeal source in mosquitoes (Picture 2).



Picture 2. Participants doing ELISA in the laboratory

4.0 Field work

Fieldwork was an important component of the training. The participants undertook mosquito sampling once weekly (Annex 1). The sampling techniques included Landing catches and Pyrethrum spray collection. The samples were brought to the laboratory and under supervision of facilitators and research assistants; each participant was allowed to do identification, parity dissections and sample preparation for ELISA for *Plasmodium* species and source of blood meal detection and for PCR. Larval sampling was also carried out to provide larvae for the insectary.

4.1 Human landing catches and Pyrethrum spray catches

The participants practiced how to do community entry, laying of spray sheets, spraying rooms with insecticide and collection of knocked down mosquitoes into petri dishes (Picture 3). Participants were also sent to some communities near Accra in the evening

and were divided into groups, with 2 participants in a group (1 indoors and 1 outdoors). They collected night biting mosquitoes off their legs with an aspirator or test tube into paper cups.



Picture 3: Participants picking mosquitoes from white sheets after praying a room with insecticide

4.2 Larval collection

The participants practiced how to identify larval breeding sites, collect mosquito larvae and identify them into species



Picture 4: Participants collecting mosquito larvae from a breeding site

5.0 Insectary management

During the first week of the training, participants were taken through the various aspects of insectary practices, management and how to use the equipment in the insectary. The participants were each provided with standard operating procedures (SOPs) for the insectary. These included feeding adult mosquitoes on sugar or blood, handling of eggs, feeding of larvae and transfer of pupae into cages. Thereafter, on daily basis, the participants were allowed to maintain the insectary colonies of *Anopheles* mosquitoes till the end of the training. This was done with the supervision of the facilitators and research assistants.

5.1 Mosquito breeding

Participants practiced daily the breeding and maintenance of mosquitoes in the insectary (Picture 4)



Picture 4: A participant feeding *Anopheles* larvae in the insectary

6.0 Certification

After the post-assessment quiz, certificates were awarded to each of the 4 participants for participating and successful completion of the two month training course.

7.0 Assessment of participants

During the pre-assessment of participants, 2 of the participants scored above 50%. However, the post-assessment indicated that all 4 participants scored 80% or higher.

8.0 Evaluation of the course

In evaluating the training course, all 4 participants rated the course as excellent. The contents, delivery and laboratory aspects of the course were also rated as excellent.

9.0 Conclusions and recommendations

In general, the intermediate level entomology training course was successful. We had to make some changes to the programme in the first week based on the results of the pre-assessment quiz. Although, some of the participants initially found it difficult to cope with the course, their level of enthusiasm was very much commendable and at the end of the course, we were satisfied that they had acquired much knowledge and skills in entomological techniques. We recommend that all the participants should as soon as possible be given the necessary support and logistics to enable them put what they have learnt into practice. We also recommend a follow-up visit by facilitators to assist and help start off the participants in their respective laboratories in Liberia.

Annex 1

PROGRAMME FOR NINE-WEEK TRAINING IN INTEGRATED VECTOR MANAGEMENT.

WEEK 1 23 RD -28 TH AUGUST 2010	0800 – 1030H	1030-1130H	1130 – 1300H	1300-1400H	1400 – 1700H
DAY 1 MONDAY	ORIENTATION	FAMILARIAZATION TOUR OF NMIMR	FAMILARIAZATION TOUR OF NMIMR	Lunch	Entomology video
DAY 2 TUESDAY	Course objectives Vector Biology -Life cycle and the significance of each stage for malaria control	Diversity of malaria vectors - <i>Anopheles gambiae</i> species complex -Ecological factors affecting species distribution	Larval sampling and processing • Larval habitats and conditions affecting adult production	Lunch	Adult sampling and processing- HLC, PSC, EXIT traps and Outdoor collections Insectary management
DAY 3 WEDNESDAY	Free-Participants went to Bank to cash money	Vector Incrimination-factors that implicates a mosquito as a vector		Lunch	Entomology video
DAY 4 THURSDAY	RECAP: Life cycle of mosquitoes, larval sampling and procession		RECAP: Adult sampling and processing	Lunch	Field trip to carry out Human Landing Catches and Pyrethrum Spray collection
DAY 5 FRIDAY	Return from night collection	Rest and prepare for sample processing		Sample processing: Sorting, species identification	
DAY 6 SATURDAY	Handouts of lectures for private reading	Handouts of lectures for private reading		Handouts of lectures for private reading	

WEEK 2 30 TH AUG-4 TH SEPT 2010	0800 – 1030H	1030-1130H	1130 – 1300H	1300-1400H	1400 – 1700H
DAY 1 MONDAY	Insectary practice and management		ELISA principles <ul style="list-style-type: none"> • Antibody-Antigen reaction 	Lunch	ELISA principles <ul style="list-style-type: none"> • Enzyme - substrate reaction • ELISA practice-pipetting
DAY 2 TUESDAY	Insectary practice and management	ELISA practice-pipetting		Lunch	Mosquito identification ELISA practice-pipetting
DAY 3 WEDNESDAY	Insectary practice and management	Practical demonstration of ELISA-sporozoite			Practical demonstration of ELISA-sporozoite
DAY 4 THURSDAY	Insectary practice and management	ELISA practice-preparation of ELISA reagents	ELISA practice-preparation of ELISA reagents	Lunch	ELISA practice- preparation of ELISA reagents
DAY 5 FRIDAY	Insectary practice and management	ELISA practice-preparation of ELISA reagents	ELISA practice-preparation of ELISA reagents	Lunch	ELISA practice- preparation of ELISA reagents
DAY 6 SATURDAY	Insectary practice and management	Entomological video	Discussion of weekly activities	Lunch	Discussion of weekly activities

WEEK 3 6 TH -10 TH SEPT 2010	0830 – 1030H	1030-1100H	1100 – 1300H	1300-1400H	1400-1700
DAY 1 MONDAY	Insectary practice and management Field sampling of mosquitoes- HLC and PSC from Odumsey-Dodowa community				
DAY 2 TUESDAY	Processing of samples collected from field <ul style="list-style-type: none"> • Sorting, identification and processing 			Lunch	Insectary management and practice
DAY 3 WEDNESDAY	Insectary practice and management	Practical: Mosquito sorting identification and processing including filing of data sheet		Lunch	Practical: Mosquito sorting identification and processing
DAY 4 THURSDAY	Insectary practice and management	Practical: Mosquito sorting identification and processing		Lunch	Practical: Mosquito sorting identification and processing
DAY 5 FRIDAY	Insectary practice and management	Practical: Mosquito sorting identification and processing		Lunch	Insectary management and practice

WEEK 4 13 TH -17 TH SEPT 2010	0800 – 1030H	1030-1130H	1130 – 1300H	1300-1400H	1400 – 1700H
DAY 1 MONDAY	Insectary practice and management Field sampling of mosquitoes- HLC and PSC from Odumsey-Dodowa community				
DAY 2 TUESDAY	Processing of samples collected from field Sorting, identification and processing			Lunch	Processing of samples collected from field Sorting, identification and processing
DAY 3 WEDNESDAY	Insectary practice and management	Processing of samples collected from field Sorting, identification and processing		Lunch	Lecture on ELISA technique
DAY 4 THURSDAY	Insectary practice and management	ELISA practicals-sample preparation, loading on ELISA plates and reading	ELISA practicals-sample preparation, loading on ELISA plates and reading	Lunch	ELISA practicals-sample preparation, loading on ELISA plates and reading
DAY 5 FRIDAY	Insectary practice and management	ELISA practicals-sample preparation, loading on ELISA plates, antibody reactions and reading	ELISA practicals-sample preparation, loading on ELISA plates and reading	Lunch	ELISA practicals-sample preparation, loading on ELISA plates and reading
DAY 6 SATURDAY	Insectary practice and management	ELISA practicals-sample preparation, loading on ELISA plates and reading			

WEEK 5 20 TH -25 TH SEPT. 2010	0800 – 1030H	1030-1130H	1130 – 1300H	1300-1400H	1400 – 1700H
DAY 1 MONDAY	ELISA practicals-sample preparation, loading on ELISA plates and reading		ELISA practicals-sample preparation, loading on ELISA plates and reading	Lunch	ELISA practicals-sample preparation, loading on ELISA plates and reading
DAY 2 Tuesday	Insectary practice and management Field sampling of mosquitoes- HLC and PSC from Odumsey-Dodowa community				
DAY 3 Wednesday	Insectary practice and management	Processing of samples collected from field Sorting, identification and processing		Lunch	Processing of samples collected from field Sorting, identification and processing
DAY 4 Thursday	Insectary practice and management	Processing of samples collected from field Sorting, identification, salivary gland dissection and parity estimation		Lunch	Processing of samples collected from field Sorting, identification, salivary gland dissection and parity estimation
DAY 5 Friday	Insectary practice and management	Processing of samples collected from field Sorting, identification, salivary gland dissection and parity estimation		Lunch	Processing of samples collected from field- Sorting, identification, salivary gland dissection and parity estimation

WEEK 6 28 TH SEPT- 2 ND OCT. 2010	0800 – 1030H	1030-1130H	1130 – 1300H	1300- 1400H	1400 – 1700H
DAY 1 Monday	Insectary practice and management Field sampling of mosquitoes- HLC and PSC from Odumsey-Dodowa community				
DAY 2 Tuesday	Insectary practice and management	Practical demonstration: WHO Insecticide susceptibility testing			Practical demonstration: Insecticide susceptibility testing and Cone assay
DAY 3 Wednesday	Insectary practice and management	Practical demonstration: WHO Insecticide susceptibility testing and Cone assay			Entomological Video
DAY 4 Thursday	Insectary practice and management	Estimation of malaria transmission parameters- man biting rates, room density etc from field data generated by participants			Estimation of malaria transmission parameters- man biting rates, room density etc from field data generated by participants
DAY 5 Friday	Insectary practice and management	RECAP- WHO Insecticide susceptibility testing and Cone assay	RECAP- WHO Insecticide susceptibility testing and Cone assay		Discussion on analysis and interpretation of transmission indices

WEEK 7 5TH – 9 TH OCT. 2010	0800 – 1030H	1030-1130H	1130 – 1300H	1300-1400H	1400 – 1700H
DAY 1 Monday	Sample processing and analysis: Dissection and estimating of transmission indices		ELISA practicals- sample preparation, loading on ELISA plates and reading	Lunch	ELISA practicals-sample preparation, loading on ELISA plates and reading
DAY 2 Tuesday	Lecture on Integrated Vector Management-principles and concept			Lunch	ELISA practicals-sample preparation, loading on ELISA plates and reading
DAY 3 Wednesday	Insectary practice and management	ELISA practicals-sample preparation for sporozoite detection and bloodmeal		Lunch	ELISA practicals-sample preparation for sporozoite detection and bloodmeal
DAY 4 Thursday	Insectary practice and management	ELISA practicals-sample preparation for sporozoite detection and bloodmeal		Lunch	Discussions on vector control options
DAY 5 Friday	Insectary practice and management	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader		Lunch	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader

WEEK 8 12 TH -16 TH OCT. 2010	0800 – 1030H	1030-1130H	1130 – 1300H	1300-1400H	1400 – 1700H
DAY 1 MONDAY	Insectary practice and management ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader		ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader	Lunch	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader
DAY 2 TUESDAY	Insectary practice and management	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader		Lunch	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader
DAY 3 WEDNESDAY	Insectary practice and management	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader		Lunch	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader
DAY 4 THURSDAY	Insectary practice and management	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader		Lunch	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader
DAY 5 FRIDAY	Insectary practice and management	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader		Lunch	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader

WEEK 9 19 TH -22 ND OCT. 2010	0800 – 1030H	1030-1130H	1130 – 1300H	1300-1400H	1400 – 1700H
DAY 1 MONDAY	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader			Lunch	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader
DAY 2 TUESDAY	Insectary practice and management	ELISA for bloodmeal detection-sample preparation, loading on plates and readings from ELISA plate reader		Lunch	ELISA for bloodmeal detection-sample preparation, loading on plates and readings from ELISA plate reader
DAY 3 WEDNESDAY	ELISA for bloodmeal detection-sample preparation, loading on plates and readings from ELISA plate reader	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader		Lunch	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader
DAY 4 THURSDAY	ELISA for bloodmeal detection-sample preparation, loading on plates and readings from ELISA plate reader	ELISA for sporozoite detection-sample preparation, loading on plates and readings from ELISA plate reader		Lunch	General Discussions on lectures and practicals
DAY 5 FRIDAY	Post-Assessment	Certificate presentation ceremony Group photograph		Lunch	CLOSING CEREMONY